Ecosphere

Reduction in nitrification during the early transition from conventional to organic farming practices

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Appendix S1: Supplemental Text, Tables, & Figures

Section 1: Synthetic qPCR Standard Sequences

Archaeal *amo*A Synthetic Standard Sequence

RefSeq IDs used to generate a consensus sequence for the Archaeal *amo*A consensus sequence:
JF271970.1, JF271971.1, JF271972.1, JF271973.1, JF271974.1,
JF271975.1, JF271976.1, JF271977.1, JF271978.1, JF271979.1,
JF271980.1, JF271981.1, JF271982.1, JF271983.1, JF271984.1,
JF271985.1, JN813556.1, JN813557.1, JN813558.1, JN813559.1,
JN813560.1, JN813561.1, JN813562.1, JN813563.1, JN813564.1,
JN813565.1, JN813566.1, JN813567.1, JN813568.1, KP272121.1,
KP272122.1, KP272123.1, KP272124.1, KP272125.1, KP272126.1,
KP272127.1, KP272128.1

Synthetic Archaeal *amo*A (“AOA”) Standard Sequence
ATTCTAATGGTCTGGCTTAGACGATGTACGCACTACTTATTCATAGTAGTCGTTGCAGTCAACT
CAACCCTGCTTACAATCAACGCAGGAGACTACATCTTCTACACTGACTGGGCATGGACTTCATT
TGTCGTGTTCTCAATATCACAGACATTGATGTTTGTAGTAGGTGCAACTTACTATCTGACATTT
ACAGGAGTTCCAGGAACCGCAACATACTACGCGCTTATTATGACCGTGTATACATGGGTCGCAA
AAGGCGCATGGTTTGCACTAGGTTACCCATATGACTTCATTGTTACACCAGTTTGGATACCATC
AGCAATGCTGCTTGATTTGGCATACTGGGCAACAAAGAAGAACAAGCACTCACTGATACTATTC
GGTGGTGTATTGTGTGGAATGTCACTGCCATTGTTCAACATGGTAAATCTAATGACCGTGGCTG
ATCCATTGGAGACTGCATTCAAATATCCAAGACCAACATTGCCTCCATACATGACTCCAATAGA
ACCCCAAGTGGGCAAGTTCTATAACAGTCCAGTTGCACTCGGTGCAGGCGCAGGTGCTGTATTA
ACAGTTACCTTTGCCGCTCTGGGATGTAAACTGAATACGTGGACATACAGATGGATGGCCGCAA
T

Bacterial *amo*A Synthetic Standard Sequence

RefSeq IDs used to generate a consensus sequence for the Bacterial *amo*A consensus sequence:

JF271927.1, JF271928.1, JF271929.1, JF271930.1, JF271931.1,
JF271932.1, JF271933.1, JF271934.1, JF271935.1, JF271936.1,
JF271937.1, JF271938.1, JF271939.1, JF271940.1, JF271941.1,
JF271942.1, JF271943.1, JF271944.1, JF271945.1, JF271946.1,
JF271947.1, JF271948.1, JF271949.1, JF271950.1, JF271951.1,
JF271952.1, JF271953.1, JF271954.1, JF271955.1, JF271956.1,
JF271957.1, JF271958.1, JF271959.1, JF271960.1, JF271961.1,
JF271962.1, JF271963.1, JF271964.1, JF271965.1, JF271966.1,
JF271967.1, JF271968.1, JF271969.1, JN813545.1, JN813546.1,
JN813547.1, JN813548.1, JN813549.1, JN813550.1, JN813551.1,
JN813552.1, JN813553.1, JN813554.1, JN813555.1, KP259843.1,
KP259844.1, KP259845.1, KP259846.1, KP259847.1, KP259848.1,
KP259849.1, KP259850.1, KP259851.1, KP259852.1, KP259853.1,
KP259854.1, KP259855.1, KP259856.1, KP259857.1, KP259858.1,
KP259859.1, KP259860.1, KP259861.1, KP259862.1, KP259863.1,
KP259864.1, KP259865.1, KP259866.1, KP259867.1, KP259868.1,
KP259869.1

Synthetic Bacterial *amo*A (“AOB”) Standard Sequence

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TGGGGTTTCTACTGGTGGTCACACTATCCAATTAACTTTGTAATGCCATCAACCATGATTCCAG
GTGCGTTGATGTTGGATACTATTTTATTTCTGACCCGTAACTGGCTGGTGACAGCGCTGTTTGG
TGGCGGATTCTTTGGATTATTCTTCTATCCAGGCAACTGGCCGATTTTTGGACCGACCCACTTG
CCAGTCGTAGTTGAAGGCGTATTGTTATCGATGGCTGACTACACGGGATTCCTGTATGTACGTA
CAGGTACACCTGAATATGTTCGCTTGATTGAACAAGGCTCACTGCGTACGTTTGGTGGTCATAC
CACGGTAATTGCAGCATTCTTCTCAGCGTTCGTATCGATGCTGATGTTCGTTGTATGGTGGTAC
TTTGGCAAAGTCTATTGCACCGCTTTCTACTATGTTAAAGGTAAAAGAGGTCGTATCGTATGAA
GAATGACGTTACAGCATTTGGTGAAGAAGGCTTTGCCGAGGGGA
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***nos*Z Synthetic Standard Sequence**

RefSeq IDs used to generate a consensus sequence for the *nos*Z consensus sequence:
AY425158.1, AY425160.1, AY425161.1, AY425162.1, AY425163.1,
AY425164.1, AY425165.1, AY425166.1, AY425167.1, AY425168.1,
AY425169.1, AY577558.1, AY577559.1, AY577560.1, AY577561.1,
AY577562.1, AY577563.1, AY577564.1, AY577565.1, AY577566.1,
AY577567.1, AY577568.1, AY577569.1, AY577570.1, AY577571.1,
AY577572.1, AY577573.1, AY577574.1, AY577575.1, AY577576.1,
AY577577.1

Synthetic *nos*Z ("*nos*Z") Standard Sequence
CGTTGTTCCTCGACAGCCAGGTCGTCAAGTGGAACATCGATCACGCCAAGCGCGCCTTCAAGGG
CGAGAAGGTCGATCCGATCATCCAGAAGCTCGACGTCCACTATCAGCCCGGCCACAACCACACC
TCGATGGGCCAGACCAAGGAGGCCGACGGCAAATGGCTGATCTCGCTGAACAAGTTCTCGAAGG
ACCGCTTCCTGAACGTCGGCCCGCTGAAGCCGGAGAACGACCAGCTGATCGACATCTCCGGCGA
CAAGATGAAGCTGGTGCACGACGGCCCGAGCTTCGCCGAGCCGCACGACGCCACCATCGTCCAC
CGCTCCAAGATCAACCCGATCTCGGTCTGGAACCGCGACGATCCGATGTTCGCCGATGCGGTCA
AGCAGGCCAAGGCGGACGGCGTCAACCTCGAGGCCGACTCCAAGGTGATCCGCGACGGCAACAA
GGTCCG

Section 2: Supplemental Tables

Table S1: Count of observations from the Stroud Preserve for each sampling date. Note that on 9/15/21 each of the 8 fields were sampled, but that the sample from Field 11 amplified poorly and as a result was dropped from the dataset prior to analysis with the remainder of the dataset.

Sampling Date	Nitr. & Min.	EEA	qPCR
3/4/20	8	8	8
5/5/20	8	8	
6/3/20	8	8	
7/14/20	8	8	8
8/19/20	8	8	
9/21/20	8	8	8
11/3/20	8	8	
3/10/21	8	8	8
5/10/21	8	8	
6/8/21		8	
7/14/21	8	8	8
8/11/21	8	8	
9/15/21	8	8	7
11/29/21	8	8	

Table S2: Spearman’s and Pearson’s correlations between measured soil moisture (%) and each of the measured outcomes.

		Spearman's Correlation			Pearson's Correlation			
Parameter	N *	Spearman's Rho	S statistic	p-value	Pearson's r	DF	T statistic	p-value
Net Nit.	53	0.33	16526	**0.015**	0.28	51	2.10	**0.04**
Soil NH_4^+-N	53	0.27	18056	**0.049**	0.23	51	1.68	**0.098**
Net Min.	53	-0.27	31568	**0.049**	-0.26	51	-1.90	**0.064**
Soil NO_3^--N	53	-0.33	33030	**0.016**	-0.23	51	-1.69	**0.098**
BG	53	0.17	20464	0.21	0.21	51	1.52	0.136
NAG	53	0.18	20402	0.203	0.20	51	1.49	0.144
AP	53	0.10	22238	0.46	0.15	51	1.10	0.275
NAG:BG	53	0.12	21788	0.385	0.14	51	1.01	0.32
NAG:AP	53	0.04	23784	0.769	0.00	51	-0.03	0.978
AOA	32	0.14	4710	0.454	0.17	30	0.97	0.341
AOB	32	0.07	5080	0.707	0.09	30	0.50	0.619
*nos*Z	32	0.14	4674	0.432	0.01	30	0.08	0.938

* Number of complete observed pairs between each parameter and soil moisture.

Section 3: Supplemental Figures

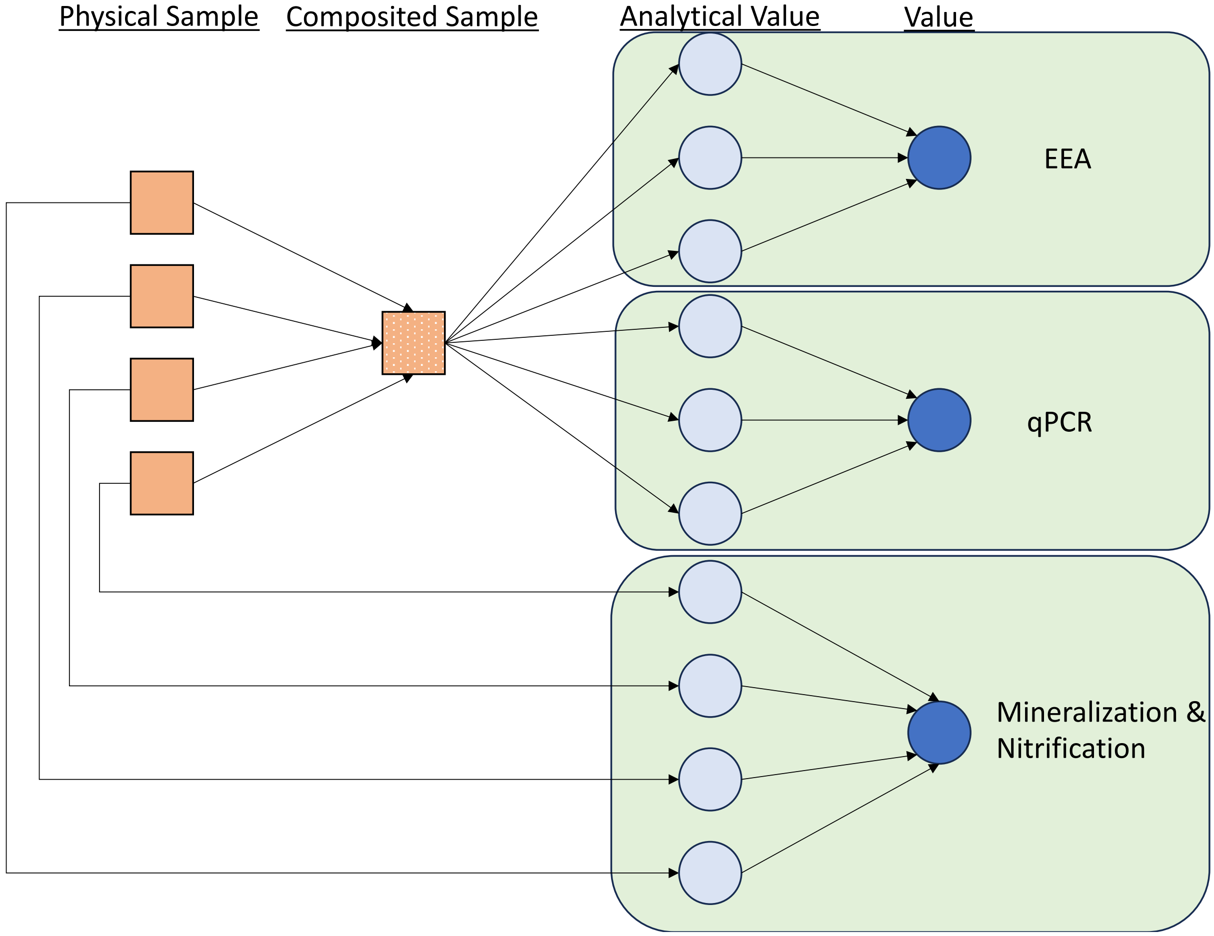

Figure S1: Diagram of sample handling and processing. Four topsoil samples (0 – 20 cm in depth) were collected from each of the 4 sub-plots within each field. Aliquots from each of the sub-plot samples were subjected to mineralization and nitrification assays; the values from these assays were averaged in order to obtain the values used in subsequent analysis. The remaining portions of the sub-plot samples were combined and homogenized into a single composited sample. Aliquots from this composited sample were then used to generate technical (analytical) replicate values for the EEA and qPCR assays. The values from the analytical replicates were averaged to obtain a single value for use in subsequent analysis.

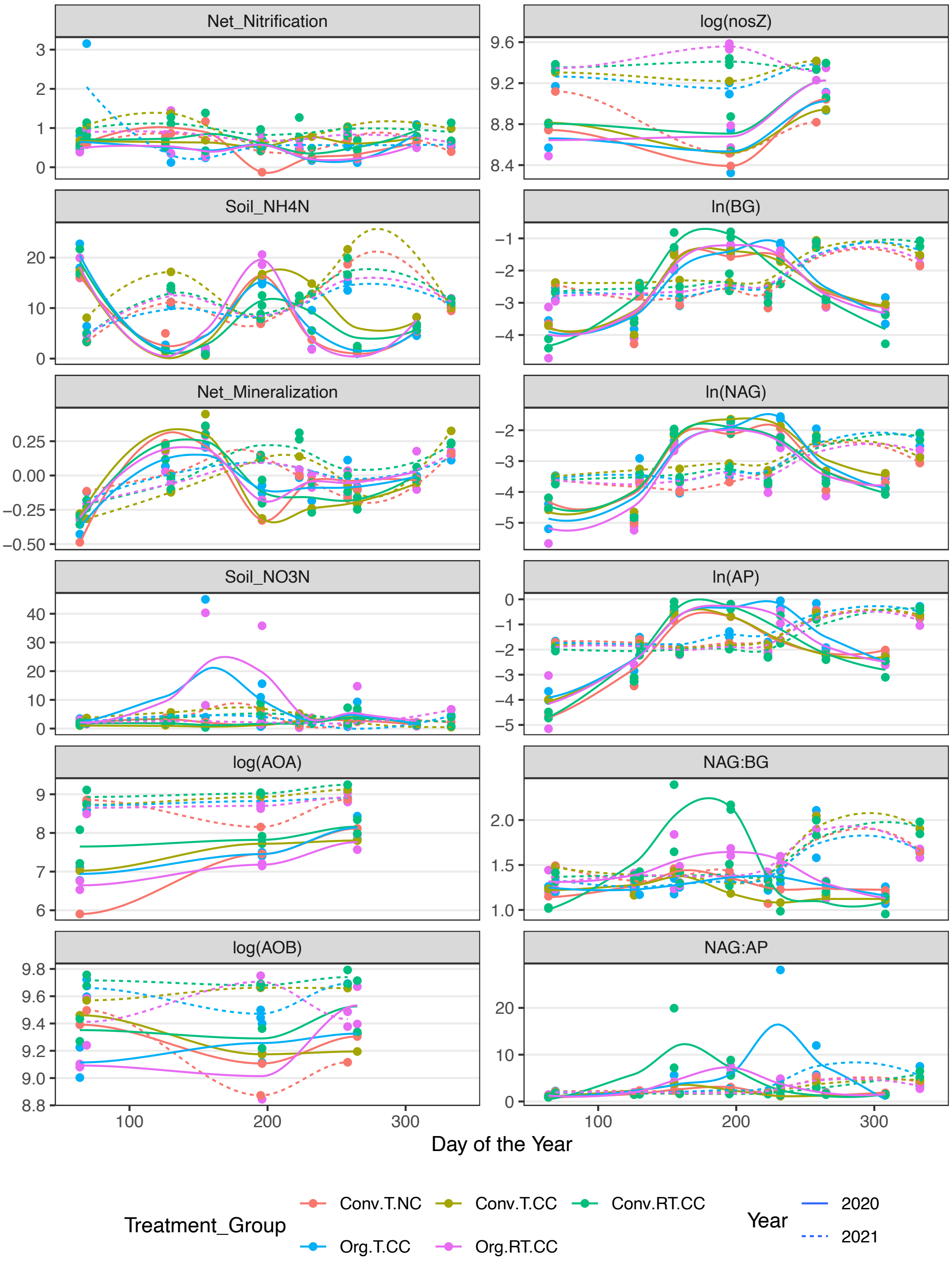

Figure S2: Seasonal patterns of the measured parameters. EEA and qPCR results were natural log transformed and log10-transformed, respectively, prior to plotting. The values of net nitrification and net mineralization were presented in mg N kg soil^{-1} d^{-1}; soil NH$_4^+$-N and NO$_3^-$-N had units of mg N kg soil^{-1}; the qPCR assays AOA, AOB, and nosZ were measured in Log10-transformed copy number g soil^{-1}; the EEA parameters BG, NAG, and AP had units of μmol of substrate g OM^{-1} hr^{-1}; the ratios NAG:BG and NAG:AP were unitless. LOESS regression was used to generate smoother curves to assist in visually interpreting the data.

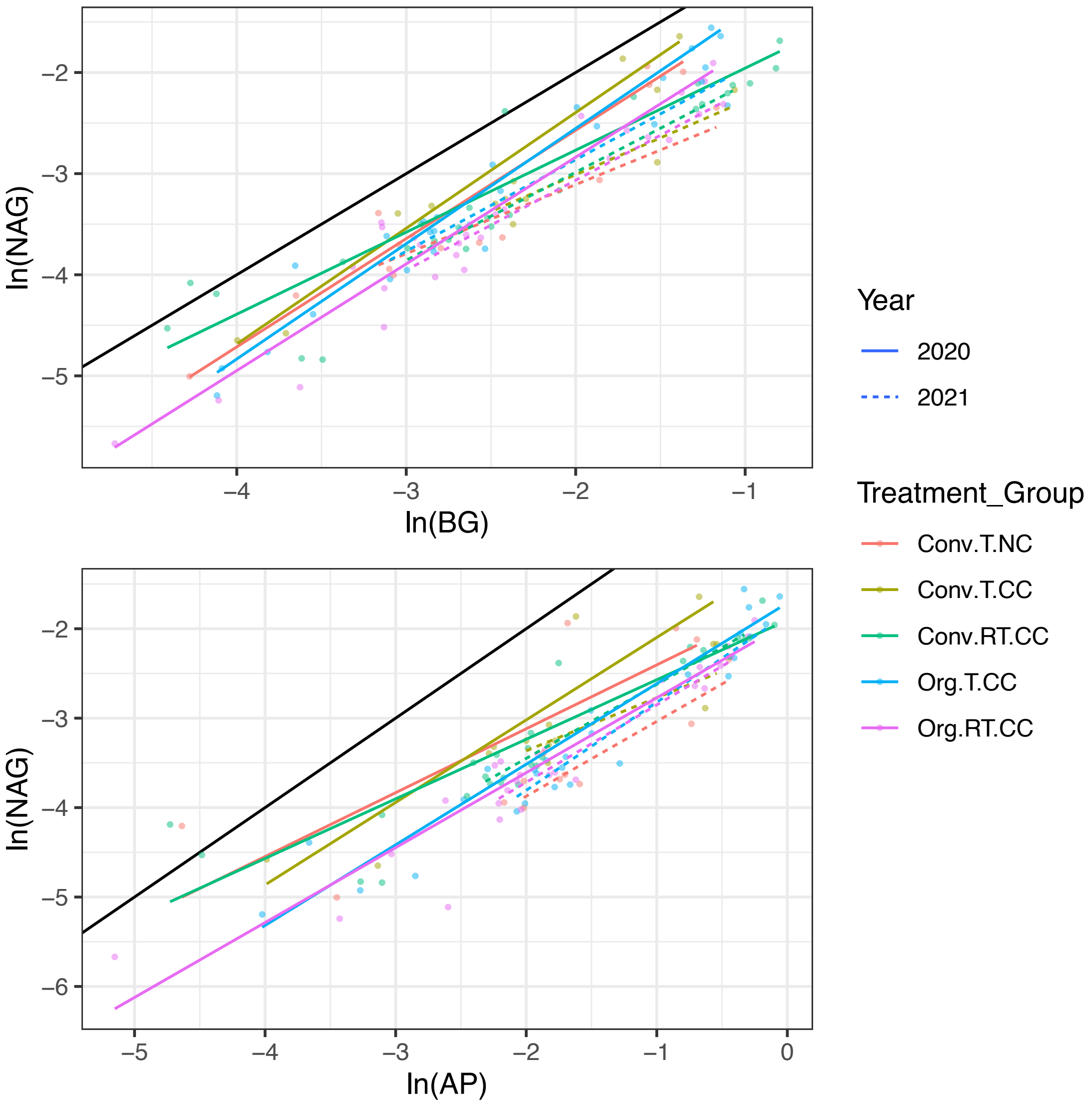

Figure S3: Ratios (NAG:BG and NAG:AP) of natural log transformed extracellular enzyme activities. The EEA parameters BG, NAG, and AP were measured with units of µmol of substrate g OM^{-1} hr^{-1}; their values were natural log transformed prior to plotting.